

## Observations on the reproductive period of the freshwater mussel *Potomida littoralis* (Unionidae)

### Observaciones sobre el periodo reproductor del mejillón de agua dulce *Potomida littoralis* (Unionidae)

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#### ABSTRACT

Data on the occurrence of gravid *Potomida littoralis* are presented from museum collections, field samplings and published literature. Gravid animals and released glochidia are recorded to occur between March and October. The smallest gravid animals were four or five years old. The length of the brooding period apparently follows a latitudinal cline. *Potomida littoralis* is most likely a tachytictic consecutive brooder with unsynchronised broods.

#### RESUMEN

Se aportan datos sobre la presencia de ejemplares grávidos de *Potomida littoralis*, a partir de colecciones de museos, muestreos de campo y bibliografía. Los reproductores y la liberación de gloquidios se produce entre marzo y octubre. Los reproductores menores eran animales de cuatro o cinco años de edad. La duración del periodo de cría sigue, en apariencia, una tendencia latitudinal. *Potomida littoralis* se reproduce en primavera y en verano, con puestas no sincronizadas.

KEY WORDS: Reproduction, Mollusca, *Potomida littoralis*, Unionidae

PALABRAS CLAVE: Reproducción, moluscos, *Potomida littoralis*, Unionidae.

#### INTRODUCTION

Freshwater mussels (Unionoida) are faced with a serious decline in both their numbers and distribution in many parts of the world (BOGAN, 1993). Reproduction biology and ecology of juvenile mussels are key factors that determine the survival abilities of freshwater mussel populations (BAUER, 2001). Several species occurring in Europe that are more seriously threatened have received considerable attention in the past decades (*Margaritifera margaritifera*:

BAUER, 1988; YOUNG, COSGROVE AND HASTIE, 2001; *Unio crassus*: HOCHWALD, 2001; *Pseudunio auricularius* (syn: *Margaritifera auricularia*): ARAUJO AND RAMOS, 2001). The biology and ecology of most other European species remain insufficiently known.

The occurrence of *Potomida littoralis* (Cuvier, 1798) (authorship of this taxon according to FALKNER, BANK AND VON PROSCHWITZ, 2001 and FALKNER, RIPKEN AND FALKNER, 2002) continues to be

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Figure 1. Orientation map for localities from Table I. F: France; E: Spain; P: Portugal; TR: Turkey; SYR: Syria; IL: Israel.

Figura 1. Mapa de las localidades de la Tabla I. F: Francia; E: España; P: Portugal; TR: Turquía; SYR: Siria; IL: Israel.

widespread in Western Europe (France, Spain, Portugal), however for other parts of the species' distribution range (Greece, Turkey, Near and Middle East, Northern Africa) no information is currently available. Data on the general biology of this species date back to the beginning of the last century. HAAS (1917) discovered that the species has 4 marsupial gills (= tetrageny). He examined the anatomy of a single male specimen from France and 11 males and 3 sterile (= non-gravid) females of *Rhombunio rothi* Bourguignat, 1863 from Jaffa/Tel Aviv, Israel (HAAS, 1920). Later, he considered this species to be a synonym of *Potomida littoralis* (HAAS, 1969). It might be noted that the material of *R. rothi* studied by Haas should still be present in the Senckenberg Museum, Frankfurt/M., Germany. Lot n. SNF 000811 contains 25 specimens preserved in alcohol, none of which are gravid. The label reads: "*Rhombunio rothi* (Bgt) - Jaffa - J. Aharoni S. 1913". HAAS (1924) later examined the condition of the marsupial gills of animals collected in Spain. He proba-

bly determined the sex of these specimens by histological studies but this is not stated explicitly in his paper. His findings however suggest that *Potomida littoralis* is dioecious.

This paper is a report on the occurrence of gravid specimens of *P. littoralis* in the river Ognon (eastern France) and on the presence of gravid mussels and glochidia of this species based on published data and museum material. The data is culled from three distinct areas within the species' distribution range.

## MATERIAL AND METHODS

Sampling localities and collection dates are given in Table I. All localities are shown on an orientation map (Fig. 1). Data on gravid specimens were obtained from examination of the following collections:

1. Collection Kinzelbach, University of Rostock, Germany (Prof. Dr. R. Kinzelbach). Reference numbers in Table II: VO-78/

Table I. List of localities.

Tabla I. Lista de localidades.

No.	Sampling locality	River or stream	River basin	Departement, Province, country
<i>France</i>				
1	Cirey-les-Belleaux , 19 km NNE of Besançon	Ognon	Rhône	Haute-Saône
2	Marnay, ca 20 km WNW of Besançon	Ognon	Rhône	Haute-Saône
3	canal near Montfrin, 20 km WSW of Avignon	Gardon	Rhône	Gard
4	Clerey, 13 km SE of Troyes	Seine	Seine	Aube
5	le Guétin, about 1,5 km upstream of confluence with Loire, 9 km SW of Nevers	Allier	Loire	Nièvre
6	Brehemont, 27 km WNW of Tours	Indre	Loire	Indre-et Loire
7	Candes-St. Martin, 50 km WNW of Tours	Vienne	Loire	Indre-et Loire
8	Chauvigny, 9 km E of Poitiers	Vienne	Loire	Vienne
<i>Spain and Portugal</i>				
9	Lake of Bañolas/Banyoles, ca 16 km NNW of Geron/Girona	endorheic		Girona
10	Acequia (ditch) de San Adrián, ca 44 km ESE of Logroño		Ebro	Navarra
11	Zaragoza	Canal Imperial de Aragón	Ebro	Zaragoza
12	Sástago, app. 56 km SE of Zaragoza	Ebro	Ebro	Zaragoza
13	near Ruidera, ca 26 km SSE of Tomelloso	Lagunas de Ruidera	Guadiana	Albacete and Ciudad Real
14	Goyán/Goján, 12 km upstream of the river mouth at La Guardia/A Guarda (at 3 different but very close sites)	Miño	Miño	Pontevedra
15	near Vila Nova de Cerveira (opposite of Goyán), ca 12 km upstream of the river mouth	Minho	Minho	Viana do Castelo, Portugal
16	Tuy/Tui, 27 km upstream of the river mouth	Miño	Miño	Pontevedra
17	Salvaterra do Miño, 39 km upstream of the river mouth	Miño	Miño	Pontevedra
<i>Turkey, Syria and Israel</i>				
18	near Samanda_i, ca 25 km SW of Hatay (Antakya)	Orontes	Orontes	Hatay, Turkey
19	(1) confluence of the two outlets of the Lake of Horns (Buhairat al-Qattina or B. al-Hims) and (2) backwater	Orontes	Orontes	Syria
20	at bridge of road Horns-Tartus	Orontes	Orontes	Syria
21	stagnant backwater in the al-Ghab ENE of Ain al-Kurum	Orontes	Orontes	Syria
22	canal at crossing of the road Ain al-Kurum to Saqalibiya (Sqeblie)	Orontes	Orontes	Syria
23	Lake Tiberias (Kinneret)		Jordan	Israel

2. Collection of recent freshwater invertebrates at the Museo Nacional de Ciencias Naturales, Madrid, Spain. Reference numbers in Table II: MNCN FW-

3. Senckenberg-Museum, Frankfurt am Main, Germany. Reference numbers in Table II: SNF-

Gills of living or preserved specimens were inspected to see whether they were filled. The developmental status of the content of the gills was not always determined and is not reported here.

Live adult mussels were gently opened by hand to approximately 1 cm. The soft parts of most museum specimens were already removed from the shell and separately stored. It is important to remember that in *Potomida littoralis* the inner and outer gills become filled with eggs. In this case they are swollen and non-transparent. Filled marsupia are cream white to light brown in live animals, while unfilled gills are semi-transparent, thin and greyish colour.

Mussels from the river Ognon at Cirey-les-Bellevaux were repeatedly inspected by hand while wading on the riverbanks. The maximum depth sampled was 1.5 m. Sampling dates are given in Table III. The mussels were painted with numbers for individual recognition and measured to the nearest 1/10 mm with a veneer calliper.

## RESULTS

Table II reports the findings from collections and field observations. All gravid specimens were encountered from April to October.

The smallest gravid specimen was 39.3 mm in length (June 6, 1990, river Miño at Goyán). The smallest gravid mussel from the Ognon measured 41.0 mm (Table III). In both cases, three, perhaps four, darkly coloured growth lines were evident on the outside of the shell. This means that the animals were in their fourth (or fifth) spring/summer period when they reproduced.

Table III reports the findings of repeated observations of marked individual mussels in one locality in eastern France. Mussel with length 55.3 mm was not found gravid in the year 2000 and became gravid in 2001. Since the animal was among the largest in that population it was most likely also sexually mature in the first year of observation. This individual may have paused from reproduction in 2000 or may have reproduced exceptionally late in that year.

Excluding animals that are less than 39.3 mm, which was the size of the smallest gravid individual, the number of mussels found gravid in the river Ognon was 33%. Similar values were found for the pooled samples from the river Orontes (33%) and the Ebro at Sástago on July 18, 1916 (30%).

## DISCUSSION

Little is known about the brood behaviour of unionoidean bivalves. In many cases inferences from circumstan-

tial observations and the lack of distinction between individual and population behaviour have prevented us from detecting the actual patterns until recently (HEARD, 1998). Specific information on the reproductive period of *Potomida littoralis* is scattered. The only published records of gravid animals are from HAAS (1917, 1920) and PRASHAD (1919).

HAAS (1917) conducted regular samplings between July 18 and September 5, 1916 in the river Ebro at Sástago. He found individuals carrying eggs and larvae at the beginning of this period and he recognized freshly metamorphosed mussels on the last date. However, Haas obviously failed to account for the presence of gravid animals in a sample he had taken the year before (September 7-8, 1915; Haas 1916). A small proportion of those mussels, still preserved in the Senckenberg Museum, are gravid (Table II). For the other samples carried out in 1916 (Sástago: August 15, 22, 29, September 5; Gallur, ca 42 km NW of Zaragoza: September 3) Haas did not give the number of *P. littoralis* collected. It remains unclear whether he inspected them for gravidity at all.

PRASHAD (1919) made observations on 3 specimens of *Unio (Rhombunio) semirugatus* collected in October, 1912, in Lake Tiberias (Sea of Galilee). The species name is considered by HAAS (1969) to be a synonym of *Potomida littoralis*. One mussel was gravid in the sample of Prashad. Apparently, it contained glochidia only in the outer gills. In fact, Prashad regarded the outer gills as the only structure having marsupial function (= ectobranchy). Apart from the presence of larvae, Prashad based his identification of marsupial gills on the spacing of the gill septa. HAAS (1924) expressed doubts about the validity of this interpretation since he identified four marsupial gills in *Rhombunio rothi* by histological examinations (HAAS, 1920, 1924). Considering all observations reported in Table II, it might be concluded that the specimen of Prashad was collected at the end of

Table II. Observations on the brooding behaviour of *Potomida littoralis*. The number of gravid mussels when present is given in parenthesis after the sample size. Live glochidia: data from ARAUJO, BRAGADO AND RAMOS (2000). Abbreviations. ?: number not specified.

Tabla II. Observaciones sobre el comportamiento de cría de *Potomida littoralis*. El número de ejemplares reproductores se da entre paréntesis tras el tamaño de muestra. Gloquidios vivos: datos tomados de Araujo, Bragado y Ramos (2000). Abreviaturas. ?: número sin especificar.

Month	Live glochidia	Gravid mussels	Sample size	No. of sampling locality (from Table I)	Date	Reference		
January	No <sup>1</sup>							
February	No data	No	2	18	26.2.2001	MNCN FW-681		
March	Yes <sup>2</sup>	No	21	11	5.3.1915	Haas, 1916; SMF 003052		
April	Yes	No	6	14	16.4.1991	MNCN FW-691		
		No	9	1	23.4.2000	this study, see Table III		
		Yes	10 (3)	1	29.4.2001	this study, see Table III		
May	Yes							
June	Yes	Yes	3 (1)	15	6.6.1990	MNCN FW-640		
		Yes	36 (9)	1	10/11.6.2000	this study, see Table III		
		Yes	"several" (4)	2	23.6.2000	this study		
		Yes	6 (2)	1	23.6.2000	this study, see Table III		
		No	20	7	30.6.1991	Nagel and Badino, 2001		
		No	18	6	30.6.1991	Nagel and Badino, 2001		
		No	18	6	30.6.1991	Nagel and Badino, 2001		
July	Yes	No	33	4	1.7.1991	Nagel and Badino, 2001		
		Yes	1 (1)	10	11.7.1996	MNCN FW-1157		
		No	7	14	11.7.1990	MNCN FW-645		
		Yes	"several hundred" (?) sub-sample: 47 (14)	12, at 0-0.5m depth	18.7.1916	Haas, 1917, 1920; SMF 003054; this study		
		No	5	5	21.7.1985	Nesemann and Nagel, 1989		
		No	3	6	23.7.1985	Nesemann and Nagel, 1989		
		No	4	7	24.7.1985	Nesemann and Nagel, 1989		
		No	2	8	26.7.1985	Nesemann and Nagel, 1989		
		August	Yes	Yes "many" (2) at 0-0.5 m, "some" (all ?) at 5-6 m depth		12	2.8.1916	Haas, 1917; Haas, 1920
				No	6	21	3.8.1978	VO-78/12
Yes	more than 1 (1)			22	3.8.1978	VO-78/12a		
Yes	8 (4)			23	5.8.1978	VO-78/20		
Yes	17 (6)			24 and 25	8/10.8.1978	VO-78/28; VO-78/29		
No	8			18	4.8.1988	MNCN FW-1295		
Yes	? at 0-0.5 m, ? (all ?) at 6 m depth			12	8.8.1916	Haas, 1917		
Yes	9 (2)			20	23.8.1978	VO-78/49		
No	2			14	28.8.1990	MNCN FW-652		
September	Yes			Yes	sub-sample: 35 (5)	12	7/8.9.1915	SMF 323165; this study; (Haas, 1916)
		No	42	12	8.9.1915	Haas, 1916; SMF 003053		
		No	14	13	8.9.2000	MNCN FW-1500		
		No	1	19	11.9.2002	MNCN FW-1543		
		No	10	9	15.9.1914	SMF 003049		
		No	5	14	18.9.1990	MNCN FW-654		
October	Yes <sup>3</sup>	Yes	3 (1)	23	October 1912	Prashad, 1919		
		No	17	3	5.10.1989	Nagel and Badino, 2001		
		No	19	17	7.10.1988	Nagel and Badino, 2001		
		No	1	16	24.10.1989	MNCN FW-498		
November	No data	No	2	14	6.11.1990	MNCN FW-664		
		No	1	18	6.11.1990	MNCN FW-665		
December	No							

<sup>1</sup> four dates in 1998; <sup>2</sup> from 6.3.1997; <sup>3</sup> until 9.10.1997

Table III. Observations of the reproductive status of *Potomida littoralis* in the river Ognon near Cirey-les-Belleveaux, France. From a total of 37 individually marked animals only those are listed that could be inspected at least two times. The remaining ones were not gravid on April 23, 2000 (1 specimen) or on June 10 - 11, 2000 (12 specimens). Length of specimens as measured on day of first record. No intensive sampling on June 23, 2000.

Tabla III. Observaciones del estatus reproductor de *Potomida littoralis* en el río Ognon cerca de Cirey-les-Belleveaux, Francia. De un total de 37 especímenes marcados sólo se incluyen aquellos que se observaron al menos dos veces. Los restantes no estaban en estado reproductor el 23 de abril de 2000 (1 ejemplar) o el 10-11 de junio de 2000 (12 ejemplares). Se da la longitud tomada el día de la primera observación. No se realizó un muestreo intenso el 23 de junio de 2000.

Length (mm)	Gravid			Length (mm)	Gravid		
	2000 April 23	2000 June 10/11	2001 June 23		2000 April 23	2000 June 10/11	2001 June 23
27.8	-	-	-	47.0	-	+	-
30.2	-	-	-	47.7	+	-	-
39.0	-	-	-	48.1	-	-	-
39.4	-	-	-	48.2	+	-	-
40.0	-	-	-	49.3	+	+	-
41.0	-	-	+	50.2	-	-	-
41.8	+	-	-	51.5	-	+	-
44.6	-	+	+	51.7	-	+	-
45.6	-	-	-	54.7	-	-	-
46.6	-	-	-	55.3	-	-	+
46.7	-	+	-	58.7	-	+	-
46.8	-	-	-	65.9	-	-	†

Explanation of symbols: + gravid, - not gravid, † dead, no entry: not found.

the gravidity period and had already expelled the content of the inner gills thus giving the appearance of an ectobranchous state.

Based on the observations of HAAS (1920, 1924), NAGEL (1988, p. 3, fig. 1) hypothesized that *Potomida littoralis* is a summer breeder, i.e. tachytictic like the European species of the genus *Unio*. The observations from the river Ognon (Table III) fit well into this pattern. Recently, ARAUJO, BRAGADO AND RAMOS (2000) reported on live glochidia (valves with soft body) in drift net samples from early March to beginning of October, 1997, in the Canal Imperial de Aragón that takes water from the Ebro (sampling localities near the villages of Grisén and Alagón, between Gallur and Zaragoza, see Table II). These data confirm the tachitixis but also give evidence of a very long reproductive period.

No gravid mussels were found in France in the 64 specimens sampled in July and October. Several explanations are possible for this, for example, latitude. It may be safe to assume that the reproductive season for *P. littoralis* is abbreviated at higher latitudes where periods with water temperatures favourable for the growth of food particles (bacteria, algae) are short. There is evidence that at least some unionoid species allocate only surplus energy to reproduction (HOCHWALD, 2001 and references therein). Alternative explanations are population specific and would point to a high parasite load and natural or anthropogenic habitat disturbances. All these factors can induce individuals or entire populations to suppress reproduction. This latter phenomenon was previously described for some species (*Margaritifera margaritifera*: BAUER, 1987; ROSS, 1992; *Unio crassus*: HOCHWALD,

2001; *Hyridella depressa*: WALKER, BYRNE, HICKEY AND ROPER, 2001).

The apparently very long reproductive period in the southern locations makes consecutive breeding (more than one brood in the reproductive period) of the species quite probable, but presently there is no direct evidence for this. This form of reproductive behaviour, however, seems to be widespread among unionoid mussels (HEARD, 1998).

A mean of 30% gravid mussels was found in the samples from the Ognon, one sample from the Ebro and the pooled Orontes samples (Table II). Assuming a sex ratio of 1:1 would mean that one-third of the females did not actively participate in reproduction at the times of sampling. This pattern indicates unsynchronised breeding within a population.

From the above it is concluded that *Potomida littoralis* is a tachytictic brooder

with probably consecutive unsynchronised broods. The reproductive period can span from March to October. More data are needed to determine if the length of the reproductive period follows a latitudinal cline.

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